## Heated Wax-Emulsions with Benomyl and 2,6-Dichloro-4-Nitroaniline for Control of Postharvest Decay of Peaches and Nectarines

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## ABSTRACT

Mean diameters of lesions on fruit inoculated with Monilinia fructicola or Rhizopus stolonifer and treated with heated (52 C) or unheated (24 C) dips or sprays for 10, 20, or 30 sec with 225, 450, or 900  $\mu$ g/ml of 2, 6-dichloro-4-nitroaniline (DCNA) suspended in a wax emulsion were significantly smaller than those on fruit similarly treated with DCNA suspended in water. Similar wax-benomyl treatments at 10, 33, or 100  $\mu$ g/ml were also more effective in reducing lesion diameters of Monilinia-inoculated fruit than were benomyl treatments alone. Heated DCNA treatments, whether as dips or

sprays, and with or without wax, were more effective than unheated treatments. Heat improved the effectiveness of benomyl only as a dip, a 30-sec spray (without wax), or 10-sec wax spray.

Mean per cent decay of noninoculated peaches and nectarines due to infections by *Monilinia* and *Rhizopus* was significantly less when treated for 10 sec with a heated wax spray containing 450  $\mu$ g/ml DCNA and 100  $\mu$ g/ml benomyl than when sprayed for 3 sec with the same wax-fungicide mixture unheated under commercial conditions.

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The storage and marketing life of peaches (Prunus persica [L.] Batsch) and nectarines (P. persica [L.] Batsch var. nectarina (Ait.) Maxim.) is limited by postharvest diseases. Brown rot, caused by Monilinia fructicola (Wint.) Honey and M. laxa (Aderh. & Ruhl.) and Rhizopus rot, caused by Rhizopus stolonifer (Fr.) Lind, are the most important. Less important are black mold rot (Aspergillus niger v. Teigh) and blue mold rot (Penicillium sp.) (5). In 1968, postharvest rots caused 15% and 24% losses of peaches in the New York and Chicago markets, respectively (U.S. Dep. Agr., unpublished data); and caused an estimated \$2,000,000 loss to nectarine growers and shippers in California (10).

Postharvest decay of peaches and nectarines has been reduced with fungicidal dips (4), fungicide-impregnated wrappers (2), hot water treatments (7), and combination hot water-fungicide dips (9). These methods are in limited commercial use. More widely used in western areas are commercial units which spray a wax-emulsion containing a fungicide over rotating brushes. Waxes have been used on peaches and nectarines to prevent weight loss (3) and to enhance appearance (1), but wax-fungicide applications to control postharvest rots have not been evaluated critically.

Primary factors in this study were (i) effectiveness of postharvest applications of fungicides with and without wax-emulsions; (ii) comparison of dip and spray application; and (iii) effects of temperature on decay control.

MATERIALS AND METHODS.—Freshly harvested peaches, free of injury or decay, were selected from packing sheds in the San Joaquin Valley of California. Early cultivars of fruit were used for inoculation tests because incidence of natural decay is low. Two 1-mm-deep punctures were made

on each fruit, and 1 drop of a spore suspension of *M. fructicola* or *R. stolonifer* was placed on each wound. Fruit were then incubated at 24 C under a polyethylene bag for 16 to 20 hr; defuzzed by brushing in a commercial unit with 0.03% sodium dodecylbenzenesulfonate; and then rinsed with fresh water

A total of 96 treatments of inoculated, defuzzed fruit was included with all combinations of heated (52 C) and unheated (24 C) dips and sprays, with and without wax emulsion, and with 10-, 20-, and 30-sec exposures to four concentrations of fungicide. of 2,6-dichloro-4-nitroaniline Concentrations (DCNA) were 0, 225, 450, and 900  $\mu$ g/ml. Benomyl was tested at 0, 10, 33, and 100 µg/ml. Fungicides were suspended in water or in a 15 X dilution of a paraffin-base wax emulsion (Decco Peach Wax WT-52, Pennwalt Corp., Monrovia, Ca.) consisting of 26 to 31% solids by weight, and containing 0.2 to 0.5% (w/v) sodium O-phenylphenate tetrahydrate (OPP). Dip treatment was in an insulated stainless steel tank (380 liters) in which water was circulated by pump at about 190 liters/min. Temperature was maintained ±1 C with immersed electric heating coils. Spray treatments were applied in a commercial waxing unit fitted with two flood spray nozzles placed 6 inches above a series of four rotating brushes. Spray nozzles were connected by an insulated line to the dip tank so that the same fungicide mixtures could be used for dip and spray treatments.

Five fruit constituted a treatment lot, and treatments were replicated 3 times, each with a different cultivar of peach. Because benomyl is not active against *Rhizopus* spp., test fruit inoculated with *R. stolonifer* were not treated with benomyl. Treated fruit were stored in polyethylene bags for 3

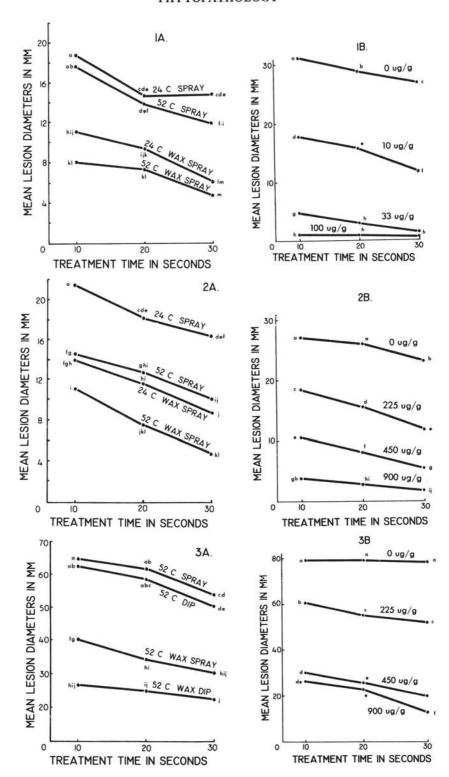


Fig. 1-3. Mean lesion diameters on peaches inoculated with 1) Monilinia fructicola, treated with benomyl; 2) M. fructicola, treated with 2,6-dichloro-4-nitroaniline (DCNA); and 3) Rhizopus stolonifer, treated with DCNA. Each point represents (A) means from 10-, 20-, or 30-sec benomyl or DCNA treatments applied by a dip method or by a spray against rotating brushes; and (B) means from 10-, 20-, or 30-sec treatments at four levels of each fungicide. Tests were replicated 3 times. Treated fruit were stored for 3 days at 4.5 C and ripened for 2 days at 21 C. Points not accompanied by the same lower case letter are significantly different at the 5% level.

days at 4.5 C, then ripened for 2 days at 21 C. The diameter of the decay lesions on each fruit was measured, and the average calculated for each test lot.

Large-scale tests with treatment lots of 100 noninoculated peaches or nectarines were conducted later in the season when the incidence of naturally occurring decay was high. Treatments were 10-sec heated and unheated dips and sprays with a mixture of 450 µg/ml DCNA and 100 µg/ml benomyl with and without a wax-emulsion. The wax-fungicide mixture also was tested as a heated or unheated spray for 3 sec, an exposure time equivalent to that in commercial units. A 90-sec dip in heated water (52 C) and a 30-sec dip in a heated suspension of 225 µg/ml DCNA, treatments currently used commercially, were included for comparison. Treated fruit were packed into commercial lugs with polyethylene liners, stored for a simulated transit and holding time of 3 days at 4.5 C and 6 to 8 days at 21 C, then examined for decay. Fruit was considered decayed if infected by Monilinia, Rhizopus, Penicillium, or Aspergillus at any stage of development.

Data were analyzed with a computer for linear regression analysis, analysis of variance, and Duncan's multiple range test. Decay percentages from the noninoculated tests were transformed to arcsins prior to analysis (8).

RESULTS.-Decay (lesion diameter) of Monilinia-inoculated fruit treated with benomyl or DCNA in combination with wax and heat .. - Lesion diameters on fruit inoculated with M. fructicola and treated with DCNA or benomyl were significantly smaller when the fungicides were suspended in a wax-emulsion rather than in water (Table 1). The greater effectiveness of wax-fungicide combinations compared to fungicide suspensions alone was evident regardless of treatment method, of temperature of application, or of specific fungicide used. Mean lesion diameters on fruit sprayed for 10, 20, or 30 sec with 24 C wax-benomyl, for example, were 11.0, 9.4, and 6.2 mm, respectively, compared to 18.8, 14.4, and 14.6 mm, respectively, on fruit sprayed with benomyl alone (Fig. 1-A). Lesion diameters on fruit correspondingly sprayed with wax-DCNA were 13.4, 11.6, and 8.6 mm, compared to 21.8, 18.4, and 16.4 mm for fruit sprayed with DCNA alone (Fig. 2-A). Mean lesion diameter of all fruit treated with wax alone was 24.2 mm, significantly less than the 30.4-mm diam of lesions on the water checks (Table 1).

Treatment temperatures also significantly affected decay. Treatments of inoculated fruit with heated DCNA suspensions, either with or without a wax emulsion, and applied either as a dip or a spray, resulted in lesions with significantly smaller diameters than treatments with unheated DCNA suspensions. Lesion diameters, for example, on fruit treated for 10, 20, or 30 sec with 52 C wax-DCNA sprays were 11.2, 7.6, and 5.0 mm, respectively, significantly less than those on fruit treated with 24 C wax-DCNA sprays (14.0, 11.6, and 8.6 mm, respectively).

Heat significantly improved the effectiveness of benomyl dip but not all spray treatments. Heated benomyl sprays were more effective than unheated sprays when the exposure time was 30 sec; and the 10-sec 52 C wax-benomyl spray (8.0 mm lesion diam) was significantly more effective than the 10-sec 24 C wax spray (11.0 mm mean lesion diam) (Fig. 1-A).

Lesion diameters decreased linearly as exposure time increased from 10 to 30 sec and, in general, no significant differences in decay could be attributed to the use of a dip or spray technique.

Decay was significantly reduced with each increment in DCNA concentration from 0 to 900  $\mu$ g/ml at each exposure time, regardless of treatment method. At 10 sec, for example, mean lesion diameters on fruit treated with 0, 225, 450, and 900  $\mu$ g/ml DCNA were 26.8, 18.6, 10.6, and 3.8 mm, respectively (Fig. 2-B). With benomyl treatments, however, each concentration was significantly different only at 10-sec exposure times. At 20 and 30 sec, there were no differences between 33 and 100  $\mu$ g/ml concentrations (Fig. 1-B).

Decay (lesion diameter) of Rhizopus-inoculated fruit treated with DCNA in combination with wax and heat.—As with fruit inoculated with M. fructicola, lesion diameters on Rhizopus-inoculated fruit were reduced significantly when DCNA was suspended in a wax-emulsion than in water alone (Table 1). Mean lesion diameter of the wax checks (78.4 mm) was not significantly different from that of the wet checks (81.4 mm). Heat had no significant effects on Rhizopus decay, regardless of treatment method.

TABLE 1. Mean lesion diameters on peaches inoculated with *Monilinia fructicola* or *Rhizopus stolonifer*, and treated with 2,6-dichloro-4-nitroaniline (DCNA) or benomyl suspended in a wax-emulsion or in water

Suspension medium	Mean lesion diameter in mm on fruit treated with indicated fungicide					
	Monilinia-inoculated			Rhizopus-inoculated		
	DCNAa	Benomyla	Checkb	DCNAa	Check	
Wax emulsion	10.66 a	8.76 a	24.2 a	35.60 a	78.4 a	
Water	15.08 b	14.88 b	30.4 b	57.64 b	81.4 a	

a Means of three replications of dip and spray treatments at 24 and 52 C with four concentrations of DCNA or benomyl
and three exposure times. Lower-case letters following means denote statistical populations at the 5% level.
b Means of three replications of dip and spray treatments at 24 and 52 C with no fungicide.

TABLE 2. Mean per cent decay of peaches and nectarines treated with 2,6-dichloro-4-nitroaniline (DCNA) and benomyl dips and sprays at 24 and 52 C (fruit was stored for 3 days at 4.5 C, and ripened 6 to 8 days at 21 C)

	Exposure time, sec	Mean per cent decay from indicated treatment temperature <sup>a</sup>		
Treatmentb		24 C	52 C	
Dry check		36.8 e	T C	
Water dip	10	39.2 e	24.1 d	
Water dip	90		10.8 bcd	
Wax dip	10	37.0 e	12.6 cd	
DCNA dip	30	36.2 e	9.4 bcd	
DCNA + benomyl dip	10	5.0 abc	4.0 abc	
DCNA + benomyl wax dip	10	3.6 abc	2.8 ab	
DCNA + benomyl wax spray	3	7.8 bcd	3.2 abc	
DCNA + benomyl wax spray	10	3.8 abc	0.6 a	

a Mean percentages of 100 fruit/cultivar infected with Monilinia fructicola, Rhizopus stolonifer, Penicillium, or Aspergillus sp. Peach cultivars Suncrest and Rio Oso, and nectarine cultivars LeGrand, Late LeGrand, and September Grand were tested. Lower-case letters following means denote statistical populations at the 5% level.

b DCNA used at 450  $\mu$ g/ml; and benomyl, at 100  $\mu$ g/ml.

Dip treatments were generally more effective than spray treatments on *Rhizopus*-inoculated fruit. The 10- and 20-sec 52 C wax-DCNA dips (26.4 and 25.0 mm mean lesion diam), for example, were more effective than the corresponding 52 C wax-DCNA sprays (mean lesion diam of 39.8 and 34.0 mm) (Fig. 3-A).

With the 30-sec treatment, each increase in DCNA concentration from 0 to 900  $\mu$ g/ml resulted in a corresponding significant decrease in *Rhizopus* lesion diameters (Fig. 3-B). At 10- and 20-sec exposure times, there were no differences between 450  $\mu$ g/ml and 900  $\mu$ g/ml DCNA.

Decay of naturally infected fruit treated with wax-fungicide dips or sprays, hot water, or hot DCNA dips.-Mean per cent decay in dry checks and in lots dipped for 10 sec in unheated water or wax (wet checks) ranged from 36.8 to 39.2% after 3 days at 4.5 C, and from 6 to 8 days at 21 C (Table 2). Lots dipped for 10 sec in heated water or heated wax developed 21.4 or 12.6% decay, respectively, levels significantly lower than in the checks. The 90-sec hot water dip and the 30-sec dip in 450 µg/ml heated DCNA, treatments that simulated commercial practice in some growing areas, resulted in 10.8 and 9.4% decay, respectively. Combination fungicide (450  $\mu$ g/ml DCNA + 100  $\mu$ g/ml benomyl) dips, with or without wax, and the wax-fungicide sprays resulted in decay levels ranging from 0.6 to 7.8%. Mean per cent decay in lots sprayed with the unheated wax-fungicide mixture for 3 sec, an exposure time equivalent to commercial practice in sheds that employ waxing units, was 7.8%. A 10-sec unheated spray and a 3-sec heated spray resulted in 3.8 and 3.2% decay, respectively, levels not significantly different from the 3-sec unheated spray. Lots treated for 10 sec with a heated wax-fungicide spray, however, developed 0.6% decay, significantly less than the decay level in the 3-sec unheated treatment. The 10-sec heated wax spray was also significantly more effective than the 30-sec hot DCNA dip or the 90-sec hot water treatment. No injury or undesirable effects on the appearance or market quality of any of the treated fruit was observed.

DISCUSSION.—In tests with artificially inoculated fruit, fungicide treatments were significantly more effective when combined with a wax-emulsion than when suspended in water. The effect of wax was independent of the specific fungicide, of the pathogen, of the method of treatment, or of the temperature.

The low levels of preservative chemical formulated into the wax concentrate may be partly responsible for the effect of wax on fungicide treatments of Monilinia. Sodium O-phenylphenate tetrahydrate (OPP), at 1,000 to 5,000  $\mu$ g/ml, is active against both Monilinia and Rhizopus (6). Approximately 133 to 300  $\mu$ g/ml OPP were present in the diluted wax emulsions in which fungicides were suspended. This level of OPP could account for the small but significant reduction of lesion diameters on fruit inoculated with Monilinia and treated with wax alone, as compared to those treated with water alone. Wax also may function as an adhesive for fungicide residues.

Heat significantly improved the effectiveness of all DCNA treatments and of some benomyl treatments against *Monilinia*. Fruit dipped in heated DCNA suspensions have higher residues than fruit dipped in unheated suspensions (10). Thus, the effect of heat on wax-fungicide treatments may similarly be attributed to increasing residues on heat-treated fruit.

Benomyl was more effective than DCNA for the control of brown rot; Rhizopus rot control required at least 450  $\mu$ g/ml DCNA with the 10- and 20-sec treatments. Thus, for the control of mixed Monilinia and Rhizopus infections, a combination of 33 to 100  $\mu$ g/ml benomyl and 450 to 900  $\mu$ g/ml DCNA was required. A 10-sec heated-wax spray containing 450  $\mu$ g/ml DCNA and 100  $\mu$ g/ml benomyl was more effective in reducing decay development than a 3-sec unheated wax-fungicide spray, a 90-sec hot water dip, and a 30-sec heated DCNA dip. The three latter treatments are in commercial use. Conversion of

currently used commercial waxers to provide a heated wax-fungicide spray and to extend exposure time from 3 to 10 sec would involve minor modifications of equipment.

DCNA is approved by the Food and Drug Administration for postharvest use on peaches and for temporary use on nectarines. Food and Drug Administration tolerance levels are 20  $\mu$ g/ml. Registration of benomyl is currently pending.

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